

Identity of the ailanthus webworm moth (Lepidoptera, Yponomeutidae), a complex of two species: evidence from DNA barcoding, morphology and ecology

John James Wilson¹, Jean-François Landry², Daniel H. Janzen³, Winnie Hallwachs³, Vazrick Nazari¹, Mehrdad Hajibabaei¹, Paul D. N. Hebert¹

1 Department of Integrative Biology, University of Guelph, 50 Stone Road East, Guelph, ON, N1G 2W1, Canada **2** Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON, K1A 0C6, Canada **3** Department of Biology, University of Pennsylvania, Philadelphia, PA, 19104, USA

Corresponding author: John James Wilson (jwilso04@uoguelph.ca)

Academic editor: E. van Nieuwerkerken | Received 2 February 2010 | Accepted 28 February 2010 | Published 17 May 2010

Citation: Wilson JJ et al. (2010) Identity of the ailanthus webworm moth (Lepidoptera: Yponomeutidae), a complex of two species: evidence from DNA barcoding, morphology and ecology. ZooKeys 46: 41–60. doi: 10.3897/zookeys.46.406

Abstract

During extensive ongoing campaigns to inventory moths of North America and Area de Conservacion Guanacaste (ACG), northwestern Costa Rica, we discovered that morphologically similar yponomeutid moths were assigned two different names, *Atteva ergatica* Walsingham in Costa Rica and *A. punctella* (Stoll) in North America, but had identical DNA barcodes. Combining DNA barcoding, morphology and food plant records also revealed a complex of two sympatric species that are diagnosable by their DNA barcodes and their facies in Costa Rica. However, neither of the names could be correctly applied to either species, as *A. ergatica* is a junior synonym and *A. punctella* a junior homonym. By linking our specimens to type material through morphology and DNA barcoding, we determined that the ACG dry forest species, distributed from Costa Rica to southern Quebec and Ontario, should be called *A. aurea*, whereas the similar and marginally sympatric ACG rain forest species found in Central America should be called *A. pustulella*. Neotypes are designated for *Phalaena Tinea punctella* Stoll, 1781 and *Deiopeia aurea* Fitch, 1857. *Atteva floridana* has identical barcodes to *A. aurea* and provisionally maintained as a synonym.

Keywords

DNA barcoding, integrative taxonomy, synonymy, *Atteva aurea*, *Atteva pustulella*, ACG, Costa Rica, wing pattern

Introduction

The ailanthus webworm moth is a conspicuous member of eastern North American micromoth assemblages and is commonly recorded in checklists and inventories as *Atteva punctella* (Stoll, 1781) (e.g. Heppner and Duckworth 1983; Ding et al. 2006; http://en.wikipedia.org/wiki/Ailanthus_webworm). The colloquial name refers to its association with the tree-of-heaven (*Ailanthus altissima*, Simaroubaceae), an ornamental introduced to Europe and North America from Asia. First planted in Philadelphia in 1784, the plant is now widely distributed across the United States and considered a serious invasive (Ding et al. 2006). The ailanthus webworm moth is native to the New World and its native hosts are trees in the genus *Simarouba* (Simaroubaceae). It is believed that once the expanding range of the tree-of-heaven reached southern Texas, this moth, presumably already present on native hosts, added this new host and expanded north on it (Becker 2009).

The name *Phalaena* (*Tinea*) *punctella* was recognized as a junior homonym almost immediately after its description but has been retained through several major works (Heppner and Duckworth 1983; Covell 1984; Heppner 1984). The two objective replacement names proposed were *Tinea punctella* Fabricius, 1787, and *Crameria subtilis* Hübner, 1822. The oldest valid name to replace *Phalaena punctella* is *Tinea pustulella* but this remained overlooked until recently (Heppner 2003). Over time seven more nominal taxa were synonymized under *Atteva pustulella*: *aurea* Fitch, 1857 (*Deiopeia*), *compta* Clemens, 1861 (*Poeciloptera*), *compta floridana* Neumoegen, 1891 (*Oeta*), *A. edithella* Busck, 1908, *A. exquisita* Busck, 1912, *A. ergatica* Walsingham, 1914, and *A. microsticta* Walsingham, 1914. Interestingly there were early suspicions that *A. aurea* and *A. pustulella* might represent different species, the former distributed in the United States, the latter in South America, but at the time there was insufficient material to support this view (Walsingham 1897). A recent taxonomic review of New World *Atteva* (Becker 2009) introduced several nomenclatural changes and recognized three separate species within the long-standing concept of *A. pustulella*: *A. pustulella*, *A. aurea*, and *A. floridana*. The separation of *aurea* from *pustulella* introduced by Becker was based on data presented here.

Atteva is the sole genus constituting the subfamily Attevininae within the Yponomeutidae. The group has a pantropical distribution but at least one species (*A. aurea*) now has a range that extends into the temperate zone. No consistent hypotheses regarding the relationships, placement, and ranking of Attevininae have been published but the prevalent view is that they likely form a monophyletic group within the Yponomeutidae (Kyrki 1990; Landry and Landry 1998; Dugdale et al. 1998; Regier et al. 2009).

The authors are currently involved in extensive ongoing campaigns to inventory all Lepidoptera species in North America (Hebert et al. 2009; www.lepbarcoding.org) and in Area de Conservacion Guanacaste (ACG), Costa Rica (Janzen et al. 2005; Janzen et al. 2009; Burns et al. 2007; Burns et al. 2008; Burns et al. 2009; <http://janzen.sas.upenn.edu/>). It was during the course of these inventories that the problem concerning the identity of *A. pustulella* first came to light. Incorporating DNA barcoding into

inventories has been very effective at uncovering cryptic species (Hebert et al. 2004; Janzen et al. 2005; Janzen et al. 2009; Burns et al. 2009; Smith et al. 2007; Smith et al. 2008; Floyd et al. 2009; Hausman et al. 2009) and revealing, then subsequently correcting, taxonomic issues (e.g. the identity of *Taygetis andromeda* in Janzen et al. 2009). This is the case presented here with the ailanthus webworm moth. When identical barcode sequences were recovered for *A. pustulella* in Ontario and *A. ergatica* in ACG, the initial response was to change all records of *A. ergatica* to *A. pustulella*. *Atteva ergatica* was originally described from Central America and was long thought to be a synonym of *A. pustulella* in North America. However, further sampling revealed two adjacent barcode clusters representing two distinct species within ACG and separable by their DNA barcodes, wing patterns, host plant and ecosystem. Through examination of type material and literature investigation, we discovered like Becker (2009), that the name applied to the North American ailanthus webworm was inappropriate, but also that what had been thought to be intraspecific variation in facies across the ACG dry forest and rain forest ecosystems actually represented two species.

The goal of the present study was to determine the correct names for each *Atteva* species for inclusion into our inventories. However, we also view this study as an example of successful integrative taxonomic efforts. Our conclusions were achieved through examination of *Atteva* type specimens, where available and involved a combined genitalia dissection and DNA extraction procedure (Knolke et al. 2005) and the sequencing of mini-barcodes (Hausman et al. 2009; Rougerie et al. personal communication). Where type specimens have been lost, original illustrations were examined. We provide DNA barcode, morphological and ecological diagnostics for the two closely related species in ACG, formerly united under *A. pustulella* and recently separated again (Becker 2009).

Methods

As part of ongoing inventories in ACG (Janzen et al. 2005; Janzen et al. 2009) and Ontario (Hebert et al. 2009), specimens assigned to the genus *Atteva* were submitted to the Canadian Centre for DNA barcoding at the Biodiversity Institute of Ontario at the University of Guelph for molecular analysis. Full-length (658 bp) DNA barcodes (Hebert et al. 2003; Floyd et al. 2009) were recorded for these specimens with standard protocols (www.dnabarcoding.ca).

Reared wild-caught specimens (see Janzen et al. 2009 for rearing methods) from the ACG caterpillar inventory are indicated by voucher codes of the form YY-SRNP-XXXXX, where the suffix contains 1–5 digits. Light-caught ACG BioLep adult specimens have the same voucher code structure, but have 6 digits in the suffix.

After the discovery that specimens identified as *A. ergatica* in ACG and *A. pustulella* in North America had identical DNA barcodes, an effort was undertaken to examine and sequence relevant type material from national collections as well as more freshly collected specimens (see specimen records in Appendix I: Dataset 1). Following the protocol of Knolke et al. (2005) DNA was extracted from old type specimens during

genitalia dissections for those which still had the abdomen intact, otherwise DNA was extracted from a leg. From these old DNA extracts, barcodes were assembled from six PCR amplicons (Hausman et al. 2009; Rougerie et al. in preparation). Wing-patterns and genitalia preparations were qualitatively examined and compared.

All sequences together with collateral information on the specimens were deposited in BOLD (www.barcodinglife.com) (Ratnasingham and Hebert 2008) in project ATTEV (Atteva of the New World). Sequences were also deposited in GenBank under accession numbers GU013569, GU692470–GU692541, HM034026–HM034136. A maximum parsimony tree was constructed in PAUP (Swofford 1998; using PAUPRat, http://users.iab.uaf.edu/~derek_sikes/software2.htm) to demonstrate the presence of diagnostic nucleotides for each species.

Results and discussion

Identical barcode sequences were recovered from so-called *pustulella* in Ontario and *A. ergatica* in ACG. Further sampling in ACG revealed two adjacent barcode clusters, one comprised of specimens entirely from the ACG rain forest and feeding on new shoot tips of *Simarouba amara* (an exclusively rain forest species) and the other of specimens entirely from ACG dry forest and feeding on new shoot tips of *Simarouba glauca* (an exclusively dry forest species). Once this was realized, it became obvious that these two species could be separated by their forewing colour pattern as well as by their barcodes. Further directed sampling then located the dry forest species feeding on both species of *Simarouba* in the several-km-wide dry forest-rain forest intergrade, in some cases on the same plant of *S. amara* side-by-side with the rain forest species. Yet more sampling further into the rain forest ecosystem (to the east of the dry forest) revealed that the dry forest species has now moved at least 20 km into the ACG rain forest ecosystem of *S. amara*, in open pastures and field edges, and under more xeric conditions than would have been the case were the site still covered with original rain forest. In this circumstance, the rain forest species is at present much more abundant than is the dry forest species.

Our conclusion that the ACG specimens comprise two species is based on concordance (Avice and Ball 1990) between morphological, molecular and ecological characters that are diagnostic (Cracraft 1983) for each population, providing evidence for a hypothesis of genetic isolation. There was no divergence in the genitalia among the species examined. This lack of a genital difference is consistent with previous morphological studies of this group (Becker 2009). Based on congruent wing patterns of the *A. punctella* type image (Figure 2A) and the wings of our rain forest cluster, we conclude that the correct name for the rain forest species is *A. pustulella*, which also fits with its rain forest distribution in the Neotropics (Becker 2009). Based on congruent wing patterns and identical DNA barcodes with the type specimens of *A. ergatica* and *A. edithella* (Figure 1), both synonymized under *A. aurea* (Becker 2009), we conclude that the ACG dry forest species, which extends north to southeastern Canada, should

be called *A. aurea* (Figure 2). The two correct names along with synonyms and diagnostic characteristics are listed below, along with two other *Atteva* species included in our inventories.

Interestingly, *A. aurea* is more proximate in barcode to *A. hysginiella* than *A. punctella* (Table 1; Figure 1). *Atteva hysginiella* is geographically disjunct as a Galapagos Islands endemic (Figure 2), strikingly different in coloration, and uses a different host plant. Although intraspecific distances (Table 1) may seem high within these species compared to values reported in other studies (e.g. Janzen et al. 2009), we observed no relationship between genetic distance and geographic distance or morphological differences (Figure 1). Short sequence lengths obtained from older specimens (see specimen records in additional file 1: Datasheet 1) can make phenetic distances seem larger than if a full barcode sequence (658bp) were available for comparison. This is a problem seen when using percentage phenetic distances but it is clear from the maximum parsimony analysis (Figure 1) and the consensus barcodes shown in Table 1, that each species possesses a cohesive cluster of haplotypes with few polymorphisms, and diagnostic characters are present which separate all species.

***Atteva pustulella* (Fabricius)**

Phalaena Tinea punctella Stoll, 1781: 164. Type locality: Costa Rica by neotype designated here. A primary junior homonym of *Phalaena punctella* Linnaeus, 1761. *Tinea pastulella* Fabricius, 1787: 241. Objective replacement name for *Phalaena Tinea punctella* Stoll, 1781. The spelling is a printing error for *pustulella*, as is clearly evident from the description.

Tinea pustulella Fabricius, 1794: 292. Justified emendation of *T. pastulella* Fabricius, 1787.

Lithosia pustulata Fabricius, 1798: 462.

Crameria subtilis Hübner, 1822: 168. Objective replacement name for *Phalaena Tinea punctella* Stoll, 1781.

Forewings. The pattern ranging from Uruguay and Argentina northwards to Costa Rica, consists of thick black outlinings around the white spots and the orange spots reduced, with the antemedial and medial orange fasciae divided into two large blotches each (Figure 3B). No other morphological features, either in colouration, other external characters or genitalia, distinguish this species from *A. aurea*.

Habitat and food plants. *Atteva pustulella* caterpillars (400+) have been found only in the ACG rain forest ecosystem and feeding only on new shoots of *Simarouba amara*. Where the ACG rain forest intergrades with ACG dry forest, to date all caterpillars from *S. amara* have been those of *A. pustulella* (while caterpillars of *A. aurea* have been found on both *S. amara* and *S. glauca* in the intergrade zone) (Figure 2).

Ailanthus glandulosa [= *A. altissima*] in Argentina (Berg 1880: 101), *Castela erecta*, in Saint Croix, Antilles (Walsingham, 1914: 331), *C. peninsularis*, *C. polyandra*, *C.*

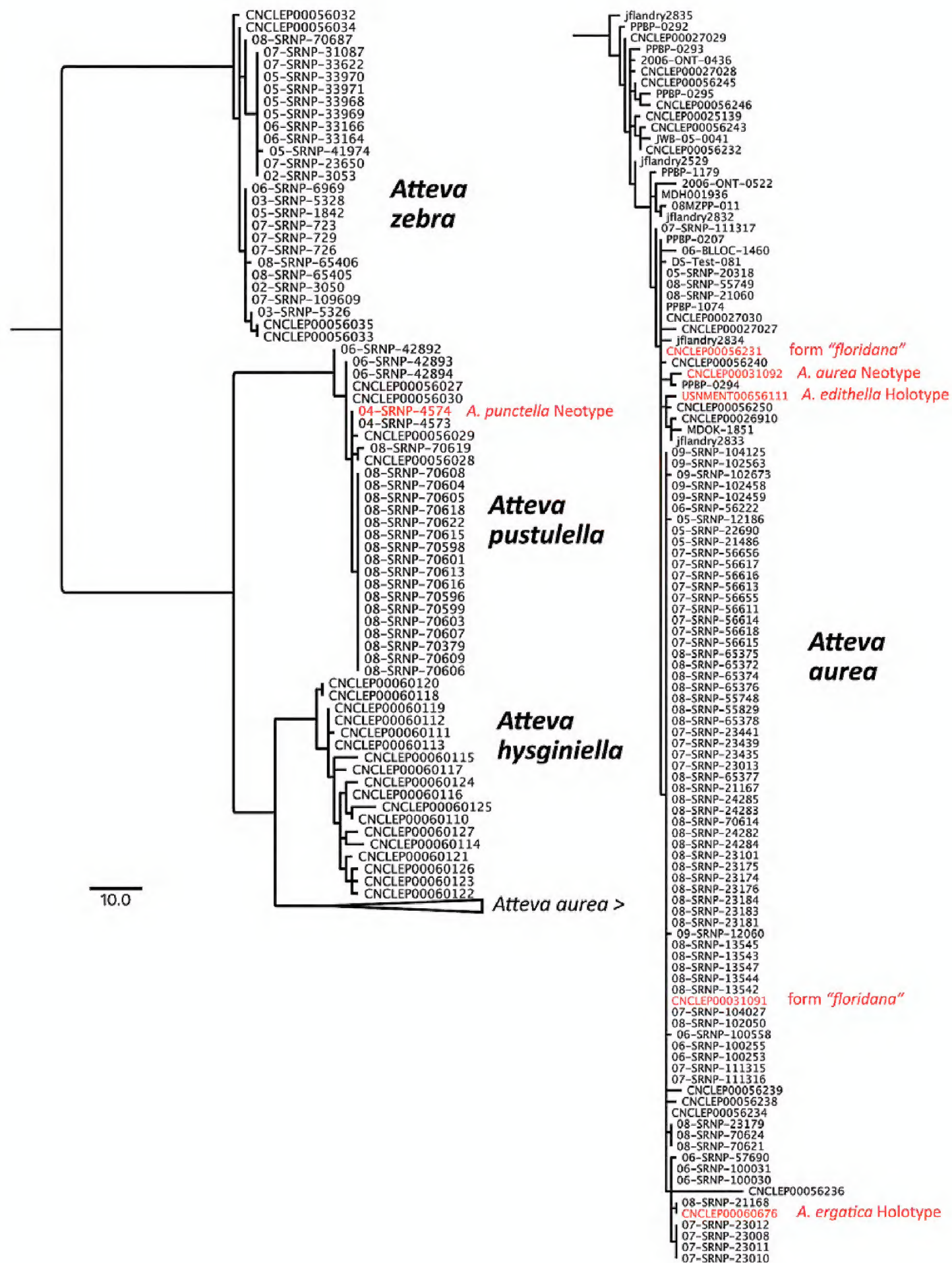


Figure 1. One of 199 most parsimonious trees found by the PAUPRat analysis of *Atteva* barcode sequences showing the presence of diagnostic nucleotide differences between the species. The scale bar shows the number of changes. Notable specimens are highlighted in red.

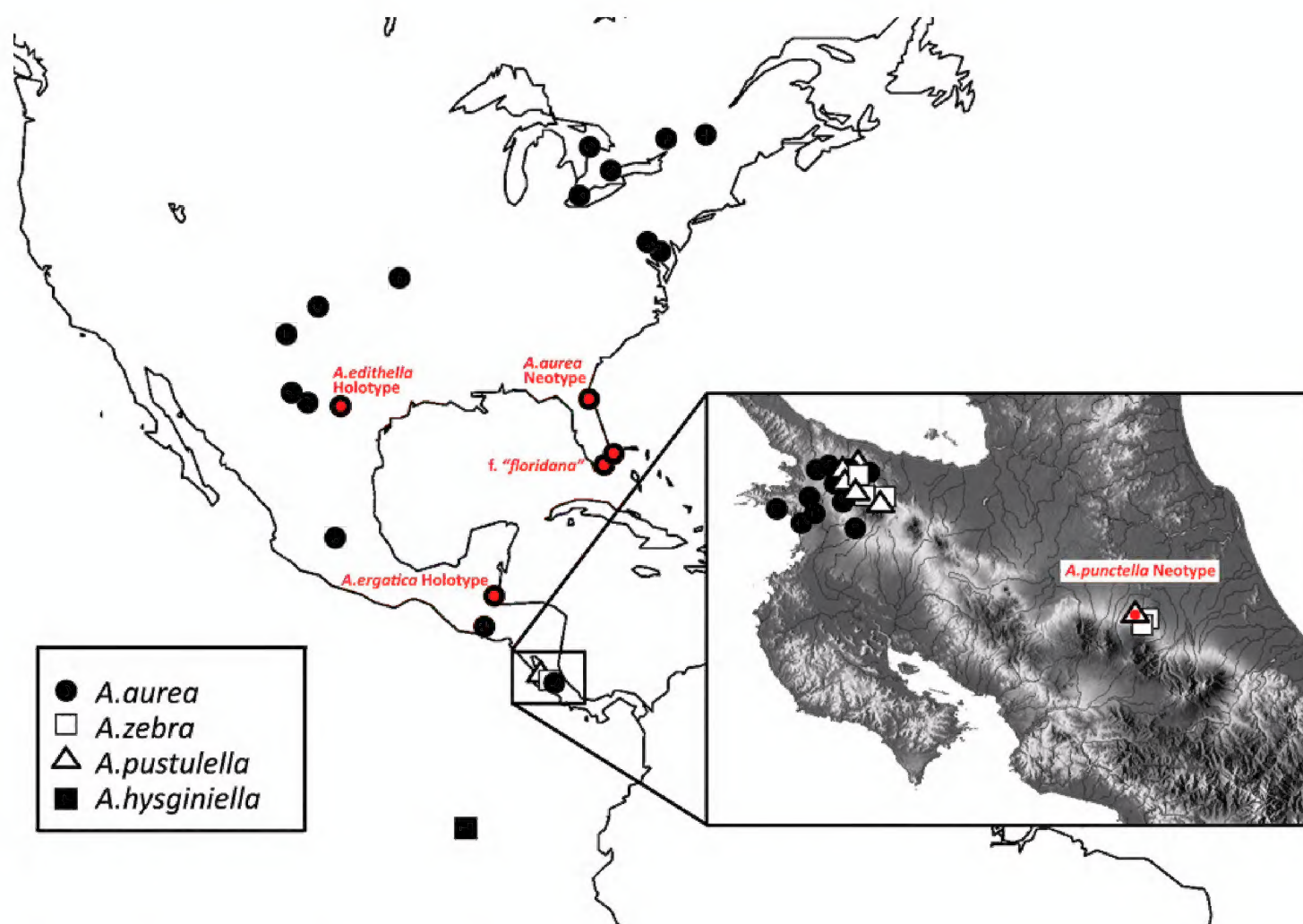


Figure 2. Map showing the distribution of *Atteva* specimens examined as part of this study. Notable specimens are highlighted in red.

emory, in the United States (Powell et al. 1973: 177). These are doubtful records for which either the host or the moth species may be misidentified (Becker 2009).

Distribution. This species ranges from Costa Rica, where it meets *aurea*, southwards to Uruguay and Argentina. It is also present in the Antilles. Becker (2009) reports several specimens from Dominica, Jamaica, Haiti and Martinique in the USNM.

Neotype. *Phalaena Tinea punctella* Stoll, female, here designated, deposited in USNM, labelled: "Voucher: D.H. Janzen & W. Hallwachs | DB: <http://janzen.sas.upenn.edu> | Area de Conservacion Guanacaste, | COSTA RICA. | 04-SRNP-4574 [white, printed with numbers handwritten]; "LEGS AWAY | FOR DNA" [pale yellow, printed]; genitalia slide [female symbol] | JFL 1680 [pale green, printed]; "NEO-TYPE | Phalaena Tinea | punctella | Stoll, 1781 | by J.-F. Landry 2010" [orange, partly printed, partly handwritten]. The specimen is unspread and is missing the right midleg which was removed for DNA extraction, but otherwise is in good condition. Barcode sequence under GenBank accession number HM034113.

Additional specimen data from Janzen's online database: latitude 10.90037°, longitude -85.37254°, elevation 500 m, Anabelle Cordoba leg., collection date 10 Sep 2004 as antepenultimate larval instar feeding on *Simarouba amara*, pupation 16 Sep 2004, adult emergence 25 Sep 2004, ACG voucher code 04-SRNP-4574. Photographs (lateral views of each side) are available in BOLD under the voucher code.

Table 1. DNA barcodes and the nature of intraspecific variation for four species of *Atteva*.

Species	Number of sequences	Number of unique haplotypes	Mean intraspecific distance
<i>Atteva pustullela</i>	23	7	0.19
AACATTATATTTTATTTTGGGAATTTGAAGAGGTATAGTAGGAACCTCTTTA- AGTTTATTAATTTCGAGCTGAATTAGGAAATCCTGGTTCTTTAATTGGGAATGAC- CAAATTTATAATACTATGTGCACAGCTCATGCTTTTATTATAATTTTATTAAT{A,G} GTTATACCTATTATAAATTGGAGG{A,G}TTTGGAAATTGACTTGTCCCATTAATATTAG- GAGCCCCAGATATAGCCTTTCCCCGAATAAATAACATAAGTTTTTGATTACTTC- CACCTTCTATTACCCTATTAATTGCAAGAAGTATTGTAGAAAATGGAGCAGGTACAG- GATGAACTGTATACCCACCTTTATCTTCCAATATTGCTCATGGAGGTGGTTCAGTT- GATTTAGCTATTTTCTCTTCATTTAGCTGGAATTTTCATCTATTTTAGGAGCTATTA- ATTTTATTACTACAATTATTAATATACGAAGTAATGGTATAAATTTTGATCAAATACCTT- TATTTGTTTGAGCTGTAGGAATTACTGCTCTTTTATTATTATTATCTTTACCAGTATTAG- CAGGGGCTATTACTATACTTTTAACTGATCGAAATTTAAATACTTCATTTTTTGACCCAG- CAGGTGGTGG{A,G}GATCCAATTTTATACCAACATTTATTC			
<i>Atteva aurea</i>	112	44	0.56
AACATTATATTTTATTTTGGGAATTTGA{A,G}{A,G}AGGTATAGTAGGAACCTCTTTA AGTTTATTAATTTCGAGCTGAATTAGGAAATCCTGG{C,T}TC{C,T}TTAATTGG{C,T} AATGACCAAATTTACAATACTATTGTTACAGCTCATGCTTTTATTATAATTTT-T- TATAGTTATACCTATTATAAATTGGAGGATTTCGGAAATTGACTTGTTCCATTA- ATATTAGGAGCTCCAGATATAGCTTTCCC{A,C,T}CGAATAAATAATATAAGTTT{C,T} TGACTACT{C,T}CCACCTTCTATTACCTTATTAATTGCAAGAAGAATTGTAGAAAAT- GGAGCAGGTACAGGATGAACTGTATACCCACCTTTATCATCTAATATTGCT- CACGGAGGTAGTTCAGTTGATTTAGCTATTTTCTTTACATTTAGC{C,T} GGAATTTTCATCTATTTTAGG{A,G}GCTATTAATTTTATTACTACAATTATTAATATAC- GAAGTAATGGTATAAATTTTGATCAAATACCTTTATTTGTTTGAGCTGTAGGAATTACT- GCTCTTTTATTATTATTATCTTTACCAGTTTTAGC{A,G}GGAGCTATTACTATACTTTTA ACAGATCGAAATTTAAATACTTCATTTTTTGACCC{A,G}GCTGGTGG{G,T}GG{A,G}{A,G} A{C,T}CCAATTTT{A,G}TA{C,T}CAACATTT{A,G}TTT			
<i>Atteva hyginiella</i>	27	15	0.75
AACATTATATTTTATTTTGGGAATTTGAAGAGGTATAGTAGGAACCTCTTTA- AGTTTATTAATTTCGAGCTGAATTAGGAAA{C,T}CC{C,T}GGTTCTTTAATTGG{C,T} AACGATCAAATTTA{C,T}AATACTATTGTTACAGCTCATGCTTTTATTATAATTTT-T- TATAGTTATACCAATTATAAATTGGAGGATTTGG{A,G}AATTGACTTGT{C,T}CCATTAATA TTAGGAGCTCCAGATATAGCCTT{C,T}CCTCG{A,G}ATAAATAATATAAGTTTCTGATTA CTTCCACCTTC{A,T}{A,T}TACTTTATTAATTGCAAGAAGAATT{G,C}TAGAAAAT{A,G} {A,G}AGCAGG{G,T}ACAGGATGAACTGTATACCCACCTTTATCATCTAATATTGCTCAC{ G,T}G{A,G}GGTAGTTCAGTTGATTTAGCTATTTTTC{C,T}TTACATTTAGCTGGAATTT CATCTATTTTAGG{A,G}GCTATTAATTTTATTACTACAATTATTAATATACGAAGTAATGG {C,T}ATAAATTTTGATCAAATACCTTTATTTGTTTGAGCTGTAGGAATTACTGCTCTTT- TATTATTATTATCTTTACCAGT{C,T}TTAGCAGGAGCTATTACTATACTTTTAACTGAT CGAAATTTAAATACTTCATTTTT{C,T}GA{C,T}CCAGC{A,G}GGTGG{A,G}GGAGA{C,T} CCAATTTTATA{C,T}CAACATTTATTT			

Species	Number of sequences	Number of unique haplotypes	Mean intraspecific distance
<i>Atteva zebra</i>	18	15	0.39
AACTTTATATTTTATTTTGGGAATTTGAAGAGG{A,G}ATAGTCGGTACCTCATTAAGATTTTTAATTCGAGCAGAATTAGGTAATCCTGGATTTTAAATTGGAGATGATCAAATT-TATAATACTATTGTAACAGCTCATGCTTTCATTATAATTTTAT{A,G}GTTATACCTATCATAATTGGAGGATTGGTAATTGATTAGTTCCTTTAATACTAGGAGCTCCTGATATAG-CATTCCCACGAATAAATAATAAGTTTGTGATTACTACCCCATCTCTTACTCTTTTA-ATTTCTAGAAGAATCGTTGAAAATGGAGCAGGAAGTGGATGAACAGTTTATCCACCTT-TATCTTCTAATATTGCTCATGGAGGAAGTTCTGTTGATTAGCTATTTTCTTTA-CATTTAGCCGGAATTCATCTATTTTAGGAGCTATTAATTTTATTACTACAATTAT-TAATATACGAAGTAATGGTATAAATTTTGATCAAATACCTTTATTTGTTTGAGCT-GTAGGTATTACAGCTTTACTTTTATTGTT{A,G}TCATTACCTGTTTTAGC{A,G}GGAGCTATTACTATACTTTTAACTGA{C,T}CG{A,G}AATCTTAATACTTCATTTTTT-GATCCTGCAGGTGGAGGAGACCCAATCCTTTATCAACATTTATTT			

The distance values shown are % Kimura-2-parameter distances as calculated in BOLD. The DNA barcodes for each species is directly below the name and statistics for each species. The bold-face nucleotides highlight those bases that are unique to a species, and the bracketed bases indicate the location and nature of haplotype variants.

Neotype designations. Enquiries at several major institutions (AMNH, ANSP, BMNH, NMNH, NYSM, USNM, ZMUC) failed to locate the type specimens of *A. aurea* and *A. punctella* (= *pustulella*). Similar negative results were indicated by Becker (2009) in his recent taxonomic review of *Atteva*. The primary types of the oldest available names appear to be lost. The evidence brought here strongly supports the occurrence of two distinct species long subsumed under the name *punctella*. The differentiating characters are in forewing coloration, DNA barcodes, larval host plants, and habitats, as described here under each respective species and in the ‘Results and Discussion’ section. However, wing coloration is the only character that can be used to interpret the old descriptions. The proper application of the two names was enabled retrospectively after differences in life history and barcodes were discovered and superimposed on the differences in forewing pattern.

The original figure of *Atteva punctella* appears on Plate 372 in the fourth volume of Cramer’s “*De Uitlandsche Kapellen Asia, Africa en America*” published in Amsterdam in 1780–1782 (copy in the Canadian Agriculture Library, Neatby Building, Ottawa). Following Cramer’s death in 1780, the work was completed by his associate Caspar Stoll, who authored the section containing the description of *punctella* (Stoll 1781; ICZN 1958). The original colour illustration of *punctella* shows distinctly the diagnostic thick black lining and reduced orange markings in the forewing characteristic of the rainforest species. Its provenance from Surinam, presumably in the coastal area near or around Paramaribo, in what we deem to have been a primary rainforest area leads us to attribute that name to the rainforest species encountered in ACG. However, the figure in Cramer (Figure 3A) is a very small painting on which watercolours were manually applied on individual copies of the book, so it is not possible to know how accurately

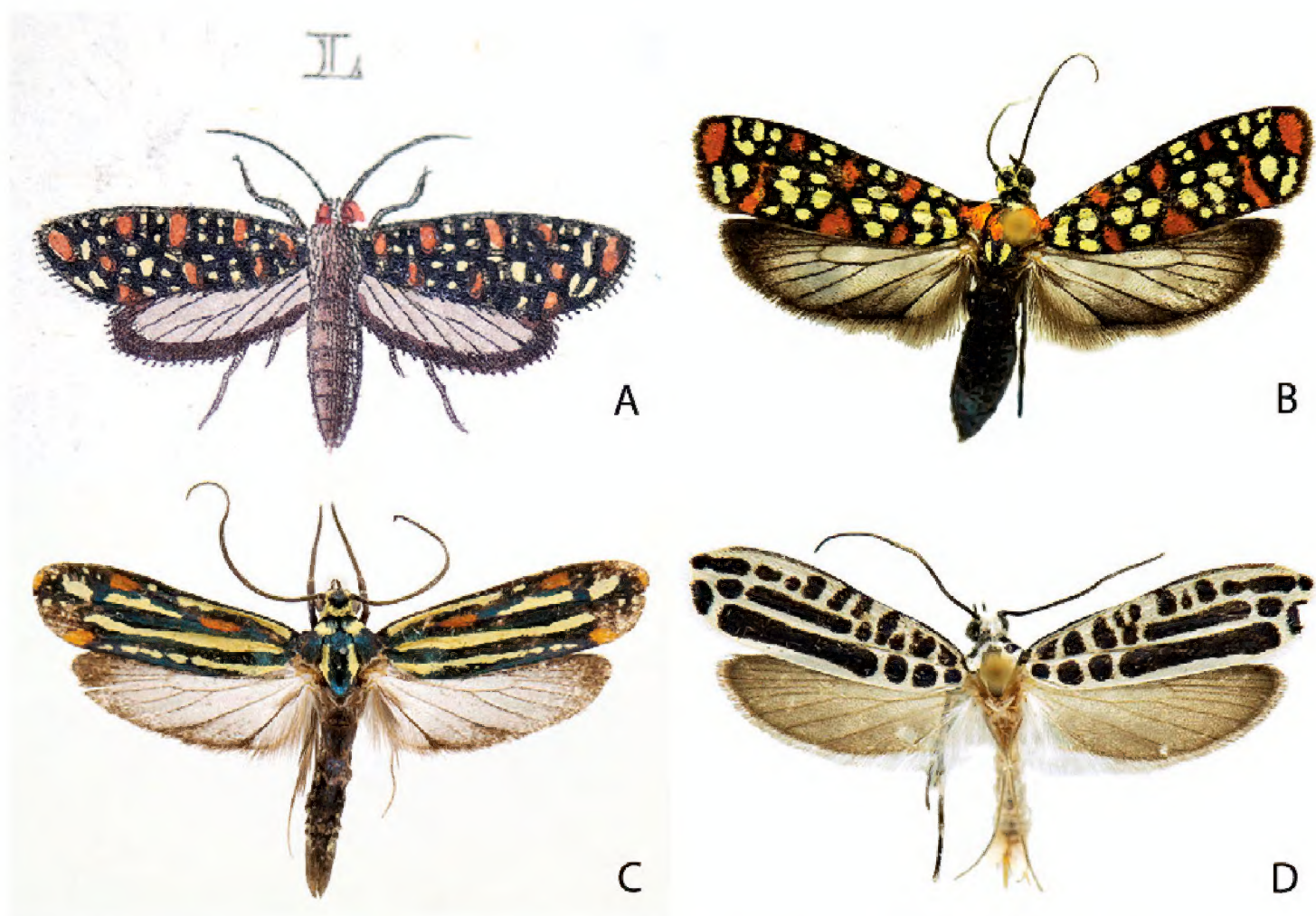


Figure 3. **A** The original figure of *Atteva punctella* from Plate 372 in Stoll (1781). The illustration is 25 mm wide in the work **B** *Phalaena Tinea punctella* Stoll (= *A. pustulella* Fabricius), specimen USNCN-CLEP00056027 (USNM) **C** *Atteva hysginiella*, specimen CNCLEP00060122 (CNC) **D** *A. zebra*, specimen CNCLEP00056033 (USNM).

the figure represented the specimen on which it was based. It was described from an unspecified number of specimens.

Although we are able to distinguish the two species on forewing pattern, there is enough variation in that character, as well as in larval host and habitat for *A. aurea* to leave open the possibility of misapplication of that name if based on these features alone. On the other hand, the barcodes are unequivocal. Therefore we deem it warranted to designate neotypes for both *punctella* Stoll and *aurea* Fitch in the interest of clarifying the taxonomic status of those nominal species and stabilizing nomenclature. The neotype was selected based on availability of a full barcode without ambiguous bases in addition to matching the forewing pattern of the original illustration of *punctella* and being from a rainforest location. While it would have been desirable to select a specimen from a locality “as nearly as practicable from the original type locality” (ICZN 1999, Art. 75.3.6), none from Surinam was available.

Atteva aurea (Fitch)

Deiopeia aurea Fitch, 1857: 486. Type locality: Marion Co., Florida, USA, by neotype designated below.

Poeciloptera compta Clemens, 1861: 526. Type locality: Texas, USA. Type not located, possibly lost. Synonymized by Berg, 1880: 100 (under *punctella*).

Oeta aurera Stretch, 1873: 240. Misspelling.

Oeta compta var. *floridana* Neumoegen, 1891: 123. Type locality: Upper Indian River, Florida, USA (USNM) [Holotype male examined (Figure 6A); genitalia on slide USNM 15942 prepared by JFL; attempt to recover DNA from abdomen failed]. Synonymized by Heppner and Duckworth, 1983: 26 (under *punctella*). Re-instated as a valid species by Becker (2009). See Remarks below.

Atteva edithella Busck, 1908: 85. Type locality: Maverick County, Texas, USA (J.D. Mitchell collector) (USNM type # 11362) [Holotype female examined (Figure 4A); genitalia on slide USNM 15940 prepared by JFL; DNA recovered from abdomen]. Synonymized by Heppner and Duckworth, 1983: 26 (under *punctella*).

Atteva exquisita Busck, 1912: 86. Type locality: Mobano, Coahuila, MEXICO (R. Muller collector, August) (USNM type # 14528) [Holotype male examined (Figure 4B); genitalia on slide USNM 92745 prepared in 1949]. Synonymized by Heppner and Duckworth, 1983: 26 (under *punctella*).

Atteva ergatica Walsingham, 1914: 328. Type locality: Rio Sarstoon, BELIZE (Blancaneaux collector) (BMNH) [Holotype female examined (Figure 4C); genitalia on slide BMNH-Microlep 31548 prepared by JFL; DNA recovered from abdomen]. Synonymized with *pustulella* by Becker (2009). The type locality is ecotone between dry forest and rain forest, the kind of habitat occupied by ACG *A. aurea* where it overlaps with the distribution of *A. pustulella*.

Atteva microsticta Walsingham, 1914: 330. Type locality: Tehuacan, Puebla, MEXICO (R. Muller collector, March 1907) (USNM) [Holotype female examined (Figure 4D); genitalia on slide USNM 92765 prepared in 1940; barcoding attempted using one leg but failed. The species name is spelled “microstigma” on the black-bordered type label in Walsingham’s handwriting.] Synonymized with *pustulella* by Becker (2009).

Forewings. The forewings are predominantly orange, with four bands of yellow spots outlined in black. Compared with *pustulella* the orange is more extended and the black outlines surrounding the white spots are thinner (Figure 5A). There is a marked amount of variation in the number and size of pale spots but in spite of this variation *aurea* is clearly separated from *pustulella* (Figures 4–6). See Remarks below.

Habitat and food plants. Caterpillars are commonly encountered in ACG dry forest feeding on *Simarouba glauca* (100+ records) and in the ecotone between dry forest and rain forest, found feeding on both *S. glauca* and *S. amara*. The *aurea* population has extended at least 20 kilometers into ACG rain forest in anthropogenic artificially more xeric conditions, feeding on *S. amara* as well. It is found to the north of Costa Rica in dry forest to semi-desert ecosystems, and north to southeastern Canada, feeding on *Ailanthus altissima*.

Distribution. In North America, distributed wherever its non-native host occurs but vagrant adults are regularly found at its northern limit in eastern Ontario and southwestern Quebec beyond the host range.

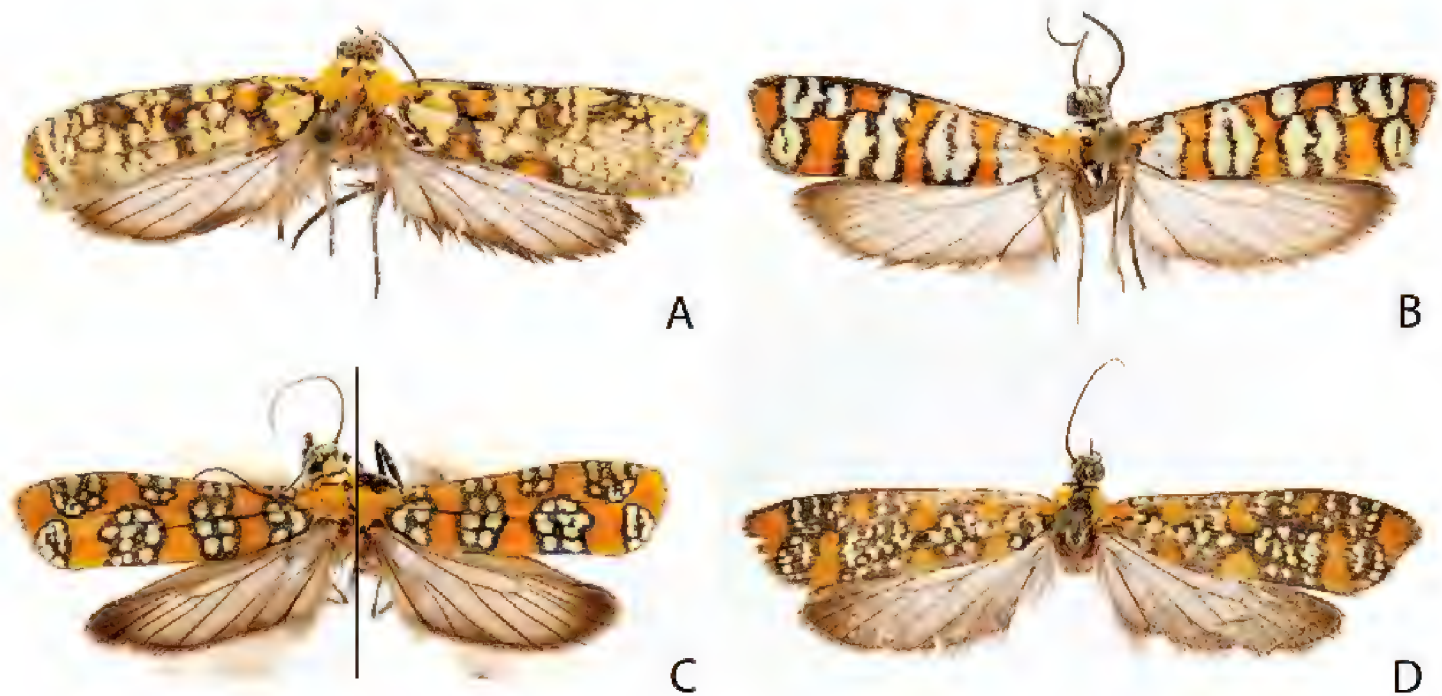


Figure 4. **A** Holotype of *A. edithella*, specimen USNMENT00656111 (USNM) **B** Holotype of *A. exquisita* from Coahuila, Mexico, specimen USNMENT00656112 (USNM) **C** Holotype of *A. ergatica*, specimen CNCLEP00060676 (BMNH); due to markedly drooped wings, two half-photos were joined to show both sides **D** Holotype of *A. microsticta*, specimen USNMENT00656110 (USNM).

Remarks. There is significant variation in forewing coloration as attested by several synonyms. More pronounced variants with reduced forewing black linings (*edithella*, *exquisita*) were described from semi-arid regions of northern Mexico and from Texas. Among barcoded specimens is an aberrant one (CNCLEP00027027 from Maryland) in which the forewing black reticulation failed to develop except asymmetrically in one small area of the right forewing (Figure 5D). That specimen was collected together with many others which displayed the normal coloration (Figure 5B–C). There is evidence that colour features including the relative extent of black reticulation and intensity of orange and pale yellow may be affected by temperature, with the black tending toward reduction at higher temperature (Powell et al. 1973).

Specimens attributable to the *floridana* have barcodes unequivocally belonging to the *aurea* array of specimens (Figure 6C–D). Attempts to barcode the old type specimen of *floridana* failed. We maintain *floridana* as a synonym of *aurea*, consistent with the traditional treatment of other authors (Heppner and Duckworth 1983; Heppner 1984, 2003; Covell 1984), but contrary to Becker (2009). Its status remains debatable, and it is possible for two species to have the same barcode. Barcode sharing is known to occur, albeit infrequently, among some close relatives in Lepidoptera (Hebert et al. 2009). The only evidence for considering *floridana* a separate species is the allegedly distinct forewing colour pattern with reduced spots (Figure 6), which is restricted to southern Florida and seems constant in sympatry with *A. aurea* (Becker 2009), whereas *A. aurea* varies considerably in forewing pattern over its wide geographic range. However, some specimens are nearly intermediate in pattern (Figure 6C) and would be questionably attributed to either *aurea* or form *floridana* from wing pattern alone. Larvae of the *floridana* form feed on *Simarouba glauca* and so overlap in host plant with *aurea*. It was reported

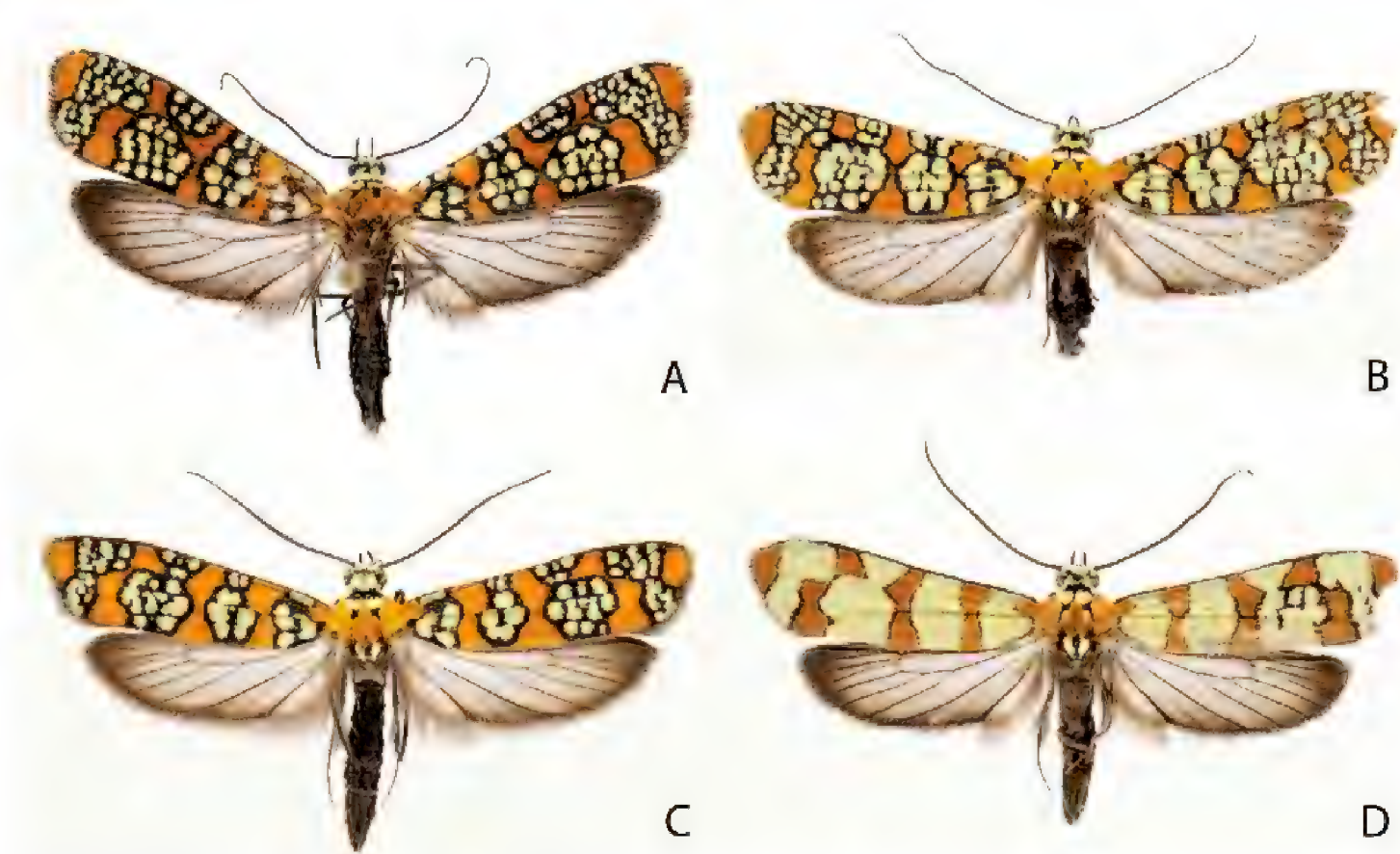


Figure 5. **A** Neotype of *Deiopeia* [= *Atteva*] *aurea*, specimen CNCLEP00031092 (CNC) **B–C** Bar-coded specimens of *A. aurea* from Maryland collected 4 Aug and 31 Jul 2006 respectively (specimens CNCLEP00027030 and CNCLEP00026910, CNC) **D** Aberrant specimen of *A. aurea* from Maryland collected 4 Aug 2006 (specimen CNCLEP00027027, CNC)

that their larvae are distinct (Walsingham 1914: 329; Becker 2009), but this contention seems to be based on incorrectly interpreting a description of the *floridana* larva (Dyar 1897) in which the two species or forms were not actually compared. The alleged difference in larvae remains unverified. Specimens seemingly of the form *floridana* examined by us were all collected early in April and May and it would be interesting to see if the form is seasonally related. Specimens of *A. aurea* were collected at various dates from May to August in the North American part of its range.

Although the type specimen of *A. aurea*, from Savannah, Georgia, appears to be lost, the description matches that of the ailanthus webworm moth and this is the oldest name applicable to North American populations of this *Atteva*. However, the original description is insufficient to determine the thickness of the forewing black lining and relative size of the orange and white spots, and there is no illustration of its type. Our attribution of that name is based on the original type locality which falls within the geographical range of the dry forest species and is far outside the range of the rain forest species. The neotype was selected based on availability of a full barcode without ambiguous bases in addition to matching the original description of the forewing pattern of *A. aurea*. While it would have been desirable to select a specimen from a locality “as nearly as practicable from the original type locality” (ICZN 1999, Art. 75.3.6), none from Georgia was available. The selected neotype is from northern Florida in a region with habitats similar to what occurs around Savannah, Georgia.



Figure 6. **A** Holotype of *Oeta* [=Atteva] *compta* var. *floridana*, specimen USNMENT00656113 (USNM) **C–D** Barcoded specimens of *A. aurea* from Dade County, Florida with a wing pattern matching or approaching that of *floridana* (all in CNC) **B** 4 Apr 2007 (CNCLEP00031090) **C** 7 Apr 2007 (CNCLEP00031091) **D** 8 May 1990 (CNCLEP00056231).

Neotype. *Deiopeia aurea* Fitch, female, here designated, deposited in CNC, labelled: “[female symbol]” [printed]; “FLORIDA: Marion Co. | Ocala National Forest | Forest Road 88 | 3.9 Miles SE of SR 316 | Longleaf Pine Sandhills | 7 DEC 2004 MV/BL | TERHUNE S. DICKEL | Database # | CNCLEP | 00031092” [white, printed]; “Barcodes of Life Project | Leg(s) removed | DNA extracted” [blue, printed]; “NEOTYPE | *Deiopeia* | *aurea* Fitch, 1857 / by J.-F. Landry 2010” [orange, partly printed, partly handwritten]. The specimen is spread and missing the left fore- and mid-legs which were removed for DNA extraction, but otherwise is in good condition (Figure 5A). Barcode sequence under GenBank accession number HM034047.

Atteva hysginiella (Wallengren, 1861)

Amblothridia hysginiella Wallengren, 1861: 386. Type: PANAMA (NHRS)

Cydosia sylpharis Butler, 1877: [87]. Type: Albermarle Island, Galapagos, ECUADOR (BMNH). Synonymized by Meyrick, 1914: 21.

Atteva monerhythra Meyrick, 1926: 278. Type: Galapagos Islands, ECUADOR (CL Collenette collector, 31st July 1924) (BMNH). Synonymized by Landry and Landry, 1998: 33.

Forewings. The three long, slender yellowish lines on a shiny black background, along the forewings readily distinguish this species from all others in the genus (Figure 3C).

Habitat and food plants. Landry and Landry (1998) gave a detailed description of adults, immatures and life-history, with the caterpillar feeding on *Castela galapageia* (Simaroubaceae).

Distribution. Restricted to the Galapagos Islands (Figure 2). The type locality recorded as being in Panama has its origin in a labeling error (Becker 2009).

Atteva zebra Duckworth

Atteva zebra Duckworth, 1967: 71. Type: Barro Colorado Island, Canal Zone, PANAMA (WD and SS Duckworth collectors, 9th May 1964) (USNM).

Forewings. The zebra pattern makes this species readily distinct from the other species presented here (Figure 3D).

Habitat and food plants. *A. zebra* is the common webworm of shoot tips of *Simarouba amara* saplings and adult trees in ACG rain forest (n = 123) It is more abundant than *A. pustulella*, but may be found on the same individual tree with *A. pustulella* and an occasional *A. aurea* in anthropogenic rain forest habitats. It has never been found on *S. glauca* or in ACG dry forest.

Distribution. Known only from Costa Rica and Panama.

Concluding remarks

This case study demonstrates the value of combining morphological, ecological and DNA barcode information when working with similar species. *Atteva* is an example where seemingly confusing morphological and ecological patterns, can be definitively partitioned in the light of discrete data such as DNA sequences. The integration and synthesis of inventories, each one necessarily regionally focused, is facilitated by DNA barcodes, an efficiently communicated online character system. This was demonstrated by the fact that taxonomic problems surrounding the *ailanthus* webworm moth persisted in the ACG for 25 years and surfaced only recently. From the starting point of DNA barcode analyses it has been relatively straightforward to reach a taxonomic conclusion by joining taxonomic knowledge in the form of the name-bearing types with ecological and morphological information. The purported difficulty in obtaining barcodes from type material has been viewed as an obstacle to the melding of DNA barcoding information with other taxonomic information. Recent studies (Hausmann et al. 2009), including this one, show that this is not necessarily the case.

Acknowledgements

We thank Rodolphe Rougerie (Biodiversity Institute of Ontario) for primer sequences for obtaining barcodes from type specimens, Robert Footitt for allowing use of his lab at the Canadian National Collection, Vitor Becker for providing an early draft of his manuscript, ACG parataxonomists for finding and rearing the specimens from the ACG, and BOLD team (Megan Milton and Kara Layton) and CCDB laboratory staff (Suresh Naik) for technical support. Yves Bousquet and Don Lafontaine contributed to discussions on neotypes.

We thank the following individuals, curators and their institutions for specimen loans and for answering queries about types: Kevin Tuck, Natural History Museum, London, UK; Don Davis and Patricia Gentili, U.S. National Museum, Smithsonian Institution, Washington, D.C.; Jason Weintraub, Academy of Natural Sciences of Philadelphia; Tim McCabe, New York State Museum, Albany; Ole Karsholt, Zoological Museum, University of Copenhagen; Erik van Nieukerken, National Museum of Natural History, Leiden; Terry Dickel, Homestead, Florida.

This research was supported by NSF grant to D.H. Janzen DEB 0072730, Wege Foundation to Guanacaste Dry Forest Conservation, and funds and grants from NSERC and Genome Canada through the Ontario Genomics Institute to PDNH.

References

- Avice JC, Ball RM (1990) Principles of genealogical concordance in species concepts and biological taxonomy. *Oxford Surveys in Evolutionary Biology* 7: 45–67.
- Becker VO (2009) A review of the New World *Atteva* Walker moths (Yponomeutidae, Atteviinae). *Revista Brasileira de Entomologia* 53: 349–355.
- Berg C (1880) Observaciones sobre la familia Hyponomeutidae (continuacion). *Anales de La Sociedad Cientifica Argentina* 10: 99–109.
- Burns JM, Janzen DH, Hajibabaei M, Hallwachs W, Hebert, PDN (2007) DNA barcodes of closely related (but morphologically and ecologically distinct) species of skipper butterflies (Hesperiidae) can differ by only one to three nucleotides. *Journal of the Lepidopterists' Society* 61: 138–153.
- Burns JM, Janzen DH, Hajibabaei M, Hallwachs W, Hebert PDN (2008) DNA barcodes and cryptic species of skipper butterflies in the genus *Perichares* in Area de Conservacion Guanacaste, Costa Rica. *Proceedings of the National Academy of Sciences USA* 105: 6350–6355.
- Burns JM, Janzen DH, Hallwachs W, Hajibabaei M, Hebert PDN (2009) Genitalia, DNA barcodes, and life histories synonymize *Telles* with *Thracides* - a genus in which *Telles arca-laus* looks out of place (Hesperiidae: Hesperinae). *Journal of the Lepidopterists' Society* 63: 141–153.
- Busck A (1908) Descriptions of North American Tineina. *Proceedings of the Entomological Society of Washington* 9: 85–95.

- Busck A (1912) New Microlepidoptera from Mexico. Proceedings of the Entomological Society of Washington 14: 83–87.
- Butler AG (1877) On the Lepidoptera, Orthoptera, and Hemiptera collected during the visit of H.M.S. 'Peterel' to the Galapagos Islands. Proceedings of the Zoological Society of London 1877: 86–91.
- Clemens B (1861) Contributions to American Lepidopterology 7. Proceedings of the Academy of Natural Sciences, Philadelphia 1860: 522–547.
- Covell CV (1984) A field guide to the moths of Eastern North America. Houghton Mifflin, Boston, 496 pp.
- Cracraft J (1983) Species Concepts and Speciation Analysis. Current Ornithology 1: 159–187.
- Ding J, Wu Y, Zheung H, Fu W, Reardon R, Liu M (2006) Assessing potential control of the invasive plant, *Ailanthus altissima*. Biocontrol Science and Technology 16: 547–566.
- Duckworth WD (1967) A new species of *Atteva* from Central America. Proceedings of the Entomological Society of Washington 69: 69–72.
- Dugdale JS, Kristensen NP, Robinson GS, Scoble MJ (1998) The Yponomeutoidea. In: Kristensen NP (Ed) Lepidoptera, Moths and Butterflies 1. Evolution, Systematics and Biogeography. Handbook of Zoology IV, 35. De Gruyter, Berlin, 120–130.
- Dyar HG (1897) *Oeta floridana* Neumoegen. Journal of the New York Entomological Society 5: 48.
- Fabricius JC (1787) Mantissa insectorum. Proft, Copenhagen, 382 pp.
- Fabricius JC (1794) Entomologia Systematica Part 2. Proft, Copenhagen, 349 pp.
- Fabricius JC (1798) Supplementum entomologiae systematicae. Proft & Storch, Copenhagen, 572 pp.
- Fitch A (1857) Third report on the noxious and other insects of the state of New York. Transactions of the New York State Agricultural Society 16: 315–490.
- Floyd R, Wilson JJ, Hebert PDN (2009) DNA barcodes and insect biodiversity. In: Footit RG, Adler PH (Eds) Insect Biodiversity: Science and Society. Blackwell Publishing, Oxford, 417–431.
- Grote AR (1873) Contribution to a knowledge of North American moths. Bulletin of the Buffalo Society of Natural History 1: 73–94.
- Hausmann A, Hebert PDN, Mitchell A, Rougerie R, Sommerer M, Young CJ (2009) Revision of the Australian *Oenochroma vinaria* Guenée, 1858 species-complex (Lepidoptera, Geometridae, Oenochrominae): DNA barcoding reveals cryptic diversity and assesses status of type specimen without dissection. Zootaxa 2239: 1–21.
- Hebert PDN, Cywinska A, Ball SL, DeWaard JR (2003) Biological identifications through DNA barcodes. Proceedings of the Royal Society of London Series B-Biological Sciences 270: 313–321.
- Hebert PDN, deWaard JR, Landry J-F (2009) DNA barcodes for 1/1000 of the animal kingdom. Biology Letters doi: 10.1098/rsbl.2009.0848.
- Hebert PDN, Penton EH, Burns J, Janzen DH, Hallwachs W (2004) Ten species in one: DNA barcoding reveals cryptic species in the neotropical skipper butterfly, *Astrartes fulgerator*. Proceedings of the National Academy of Sciences USA 101: 14812–14817.
- Heppner JB (1984) Yponomeutidae. In: Heppner JB (Ed) Atlas of Neotropical Lepidoptera, Checklist Part I. W. Junk, The Hague, 55–56.

- Heppner JB (2003) Lepidoptera of Florida, part 1, Introduction and catalog. Arthropods of Florida and neighboring land areas, vol. 17. Florida Department of Agriculture & Consumer Services, Gainesville. 670 pp.
- Heppner JB, Duckworth WD (1983) Yponomeutidae. In: Hodges RW, Dominick T, Davis DR, Ferguson DC, Franclemont JG, Munroe EG, Powell JA (Eds) Check list of the Lepidoptera of America North of Mexico. Classey, London, 26–27.
- Holland WJ (1903) The moth book. Doubleday, Page & Co, 479 pp.
- Hübner J (1816–[1826]) Verzeichniss bekannter Schmettlinge. Augsburg, 431+72pp. (index). Pp. 161–240 [published in 1822].
- ICZN [International Commission on Zoological Nomenclature] (1958) Opinion 516. Determination under the Plenary Powers of the relative precedence to be assigned to certain works on the Order Lepidoptera (Class Insecta) published in 1775 by Pieter Cramer, Michael Denis & Ignaz Schiffermüller, Johann Christian Fabricius, Johann Casper Fuessley, and S.A. von Rottemburg respectively. Opinions and declarations rendered by the International Commission on Zoological Nomenclature 19(1): 1–44.
- ICZN [International Commission on Zoological Nomenclature] (1999) International Code of Zoological Nomenclature. Fourth Edition. The International Trust for Zoological Nomenclature, London, 306 pp.
- Janzen DH, Hajibabaei M, Burns JM, Hallwachs W, Remigio E, Hebert PDN (2005) Wedding biodiversity inventory of a large and complex Lepidoptera fauna with DNA barcoding. Philosophical Transactions of the Royal Society B-Biological Sciences 360(1462): 1835–1845.
- Janzen DH, Hallwachs W, Blandin P, Burns JM, Cadiou, J-M, Chacon, I., Dapkey T, Deans A, Epstein M, Espinoza B, Franclemont J, Haber W, Hajibabaei M, Hallwachs J, Hebert P, Gauld ID, Harvey D, Hausmann A, Kitching I, Lafontaine D, Landry J-F, Lemaire C, Miller J, Miller J, Miller L, Miller S, Montero J, Munroe E, Green S, Rawlins J, Robbins R, Rodriguez J, Rougerie R, Sharkey M, Smith A, Solis MA, Sullivan B, Thiaucourt P, Wahl D, Weller S, Whitfield J, Willmott K, Wood DM, Woodley N, Wilson JJ (2009) Integration of DNA barcoding into an ongoing inventory of complex tropical biodiversity. Molecular Ecology Resources 9: 1–25.
- Kyrki N (1990) Tentative reclassification of the Holarctic Yponomeutoidea (Lepidoptera). Nota Lepidopterologica 13: 28–42.
- Knolke S, Erlacher S, Hausmann A, Miller MA, Segerer AH (2005) A procedure for combined genitalia dissection and DNA extraction in Lepidoptera. Insect Systematics and Evolution 35: 401–409.
- Landry B, Landry J-F (1998) Yponomeutidae of the Galapagos Islands, with the description of a new species of *Prays* (Lepidoptera: Yponomeutidae). Tropical Lepidoptera 9: 31–40.
- Meyrick E (1914) Hyponomeutidae, Plutellidae, Amphitheridae. Lepidopterorum catalogus 19: 1–20.
- Meyrick E (1926) On the Microlepidoptera from the Galapagos Islands and Rapa. Transactions of the Entomological Society of London 74: 269–278.
- Neumoegen B (1891) New Rhopalocera and Heterocera. The Canadian Entomologist 23: 122–126.

- Powell JA, Comstock JA, Harbison CF (1973) Biology, geographical distribution, and status of *Atteva exquisita* (Lepidoptera: Yponomeutidae). Transactions of the San Diego Society of Natural History 17: 175–186.
- Ratnasingham S, Hebert PDN (2007) BOLD: The Barcode of Life Data System (www.dnabarcoding.org). Molecular Ecology Notes 7: 355–364.
- Regier JC, Zwick A, Cummings MP, Kawahara AY, Cho S, Weller S, Roe A, Baixeras J, Brown JW, Parr C, Davis DR, Epstein M, Hallwachs W, Hausmann A, Janzen DH, Kitching IJ, Solis MA, Yen SH, Bazinet AL, Mitter C (2009) Toward reconstructing the evolution of advanced moths and butterflies (Lepidoptera: Ditrysia): an initial molecular study. BMC Evolutionary Biology 9: 280 (doi: 10.1186/1471-2148-9-280).
- Smith MA, Wood DM, Janzen DH, Hallwachs W, Hebert PDN (2007) DNA barcodes affirm that 16 species of apparently generalist tropical parasitoid flies (Diptera, Tachinidae) are not all generalists. Proceedings of the National Academy of Sciences USA 104: 4967–4972.
- Smith MA, Rodriguez JJ, Whitfield JB, Deans AR, Janzen DH, Hallwachs W, Hebert PDN (2008) Extreme diversity of tropical parasitoid wasps exposed by iterative integration of natural history, DNA barcoding, morphology, and collections. Proceedings of the National Academy of Sciences USA 105: 12359–12364.
- Stoll C (1781) [Lepidoptera]. In: Cramer P (Ed) De Uitlandsche Kapellen IV. Baalde, Amsterdam, 91–164.
- Stretch RH (1872–1873) Illustrations of the Zygaenidae and Bombycidae of North America. Author, San Francisco, 242 pp.
- Swofford DL (1998) PAUP*: phylogenetic analysis using parsimony. Version 4.0b2a. Sinauer, Sunderland, Mass.
- Wallengren HDJ (1861) Lepidoptera. In: Fregatten Eugenies Resa, K. (Ed), Zoologia IV. Stockholm, 351–390.
- Walsingham, Lord [T. de Gray] (1897) Revision of the West-Indian Micro-Lepidoptera, with descriptions of new species. Proceedings of the Zoological Society of London 1897: 54–183.
- Walsingham, Lord [T. de Gray] (1914) Tineina, Pterophorina, Orneodina, Pyralidina and Hepialina (part). Biologia centrali-americana. Insecta. Lepidoptera-Heterocera. Vol. IV. 482 pp.
- Zeller PC (1873) Beitrage zur Kenntniss nordamerikanischen Nachtfalter, besonders der Microlepidopteren. Verhandlungen der kaiserlich-königlichen zoologisch-botanischen Gesellschaft in Wien 23: 201–334.

Appendix I

Specimen records (XLS format) of *Atteva* of the New World. File format: Microsoft Excel (1997–2003). doi: 10.3897/zookeys.46.406.app.1.ds.

Copyright notice: This dataset is made available under the Open Database License (<http://opendatacommons.org/licenses/odbl/1.0/>). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.

Citations of the datasets:

Dataset published as Appendix I:

Citation: Wilson JJ et al. (2010) Identity of the ailanthus webworm moth (Lepidoptera: Yponomeutidae), a complex of two species: evidence from DNA barcoding, morphology and ecology. DATASET. File format: Microsoft Excel (1997–2003). doi: 10.3897/zookeys.46.406.app.1.ds. ZooKeys 46: 47–66. doi: 10.3897/zookeys.46.406
